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CHANGES IN VARIOUS PROTEIN PROPERTIES OF PORK MUSCLE DURING THE SMOKING PROCESS

SUMMARY—Changes in the pH, free sulfhydryl groups, amino nitrogen content and total free amino acids of untreated, heated and heated and smoked pork longissimus dorsi muscle samples were investigated. This study demonstrated that heating and heating and smoking caused changes in the pH, free sulfhydryl and amino nitrogen content of pork samples. An interesting observation was the increase in the myofibrillar protein nitrogen fraction, pH and free sulfhydryl groups of the heated samples, and the decrease of these values in the heated and smoked samples. Results of this study indicated that smoke constituents react with the functional groups of meat proteins.

INTRODUCTION

IT HAS BEEN demonstrated that changes occur in the functional groups of meat proteins during heating. The pH of muscle tissue of various meats and poultry increased during heating (Hamm et al., 1960; Kauffman et al., 1964; Paul et al., 1966) more rapidly and to a higher level with increased temperature. Using cured hams, Cohen (1966) and Karmas et al. (1964) observed a similar change in pH upon heating. Hamm et al. (1965) observed the release of free sulfhydryl groups during the heat denaturation of meat and Bautista et al. (1961) observed a decrease in the amino nitrogen content upon heating beef muscle. Studying the total free amino acid content of raw and cooked pork, Krol (1966) and Usborne et al. (1968) observed no significant difference due to heating. The effect of the smoking process upon the functional groups of proteins has been examined by several workers. Kakō (1968) and Krylova et al. (1962) observed a decrease in pH of smoked meat. Krylova et al. (1960) indicated that the chemical activity of functional groups (amino, hydroxyl and sulfhydryl) increased as proteins became denatured during the smoking process. Phenols and polyphenols react with sulfhydryl groups and carbonyls with amino groups (Krylova et al., 1962), and interactions occur between various phenol components and individual amino acids (Kurko, 1967).

Although it has been demonstrated that heating as well as the smoking process affects protein functional groups, it was the objective of this study to determine whether smoke per se had any additional effect besides that exerted by heat during the smoking process.

EXPERIMENTAL

THE SAMPLES used in this study were prepared similarly to those reported by Randall et al. (1970). The samples for pH measurements were prepared by homogenizing 10 g of the meat sample in 100 ml of distilled water for 1 min. All pH measurements were performed with a Corning Model 12, expanding-scale pH meter.

The Sørensen method as outlined in A.O.A.C. (1965) was used to determine the amino nitrogen content. A determination of the total ninhydrin positive material was used as an estimate of the total free amino acids in the sample and is an adaptation of that used by McCain et al. (1968). The samples were prepared by the method of Tallan et al. (1954).

Ellman's reagent (1959), 5,5′-dithiobis (2-nitrobenzoic acid) (DTNB), a water-soluble disulfide for the determination of free sulfhydryl groups, was adapted for use. The reagent was prepared by adding 39.6 mg DTNB to 10 ml 95% ethanol. A 2.5-g meat sample was homogenized with 25 ml of 8 M urea for 1 min. The slurry was centrifuged at 25,000 × g for 10 min at 0°C. For analysis, this supernatant was diluted 1 to 5 with phosphate buffer (μ = 0.1, pH 8.0) and filtered through Whatman No. 2 filter paper. The colorimetric procedure was carried out as described by Ellman (1959).

The myofibrillar protein nitrogen fraction was obtained as outlined by Randall et al. (1970).

RESULTS & DISCUSSION

DATA on the changes of protein properties of untreated and treated pork loin samples are given in Table 1. The pH of the muscle tissue increased with heating and decreased with heating and smoking. A slight increase in pH values with heating was observed by Cohen (1966), Hamm et al. (1960), Kauffman et al. (1964) and Paul et al. (1966) with ham, beef, pork and rabbit muscles, respectively. Hamm (1966) has suggested that the pH changes occurring during heating of meat may be caused by charge changes or hydrogen bonding, or both, within the myofibrillar proteins. A decrease of pH on smoking meats at 20°C (Krylova et al., 1962) and at 25°C (Kakō, 1968) has been observed. The changes observed in the heated and smoked samples were probably caused by the penetration of smoke components, such as organic acids, into the meat.

Appreciable differences in the free sulfhydryl groups of untreated, heated and heated and smoked pork samples were observed (Table 1). The significant increase of sulfhydryl groups of heated pork samples was in agreement with an earlier study by Hamm et al. (1965). These workers observed a steady increase within the temperature range of 30 to 70°C and attributed this increase of free sulfhydryl groups to the unfolding of peptide chains, especially those of actomyosin. With the smokehouse conditions utilized in this study, a loss of 24% of the free sulfhydryl groups occurred in the heated and smoked pork samples. Krylova et al. (1962) observed a 60% decrease in the free sulfhydryl groups of smoked beef. The decrease observed was probably due to the formation of complexes between smoke components and free sulfhydryl groups, since Krylova et al. (1962) noted that the phenolic fraction of smoke exerted the greatest effect upon these groups.

Table 1—Effect of heating and heating and smoking on protein properties of pork samples.[1,2]

Variables	State of muscle		
	Untreated	Heated	Heated-smoked
pH	5.31	5.48	4.95**
Free sulfhydryl groups (μmoles/g protein)	91.87	120.37	69.81**
Amino nitrogen (mg/g protein)	9.05**	7.06	6.57
Ninhydrin positive material (μmoles/g protein)	526.67	559.67	540.30
Ninhydrin positive material (μmoles/g material)	179.90*	259.70	229.90

*P < .05, **P < .01.

[1]Smokehouse condition: 60°C (140°F), 45%R.H., 2.25 hr.
[2]Means underlined by the same line do not differ significantly.

There was an appreciable change in the amino nitrogen content of the heated and heated and smoked pork samples (Table 1), with the majority of the decrease being due to heat effects. Upon heating beef longissimus dorsi muscle to 65°C, Bautista et al. (1961) observed a similar decrease. Kihara (1962) obtained a slight increase in the amino nitrogen content (mg/g meat) of smoked poultry and pork as did Kido et al. (1967) with smoked herring, but this increase was probably a reflection of the weight lost during smoking rather than an actual increase in the amino nitrogen content.

McCain et al. (1968) stated that the determination of the total ninhydrin-positive material may be used as an estimation of the total free amino acids in the sample. By this determination, no appreciable changes were observed between any of the samples (Table 1) when calculated on a protein basis. Usborne et al. (1968) observed a noticeable increase of total free amino acids during heating of pork. de Abreu et al. (1962) had a similar increase during smoke drying of sausages and Kido et al. (1967) found a 50% increase of total extractable amino acids of smoked herring based upon the wet weight of the sample. By calculating the ninhydrin-positive material on a wet weight basis (Table 1), a significant increase ($P < .05$) was observed between the treated and untreated samples.

It appeared that a relationship existed between the pH, free sulfhydryl groups and the myofibrillar protein nitrogen fraction (Table 2). An increase was observed in all 3 values from the heated pork samples, whereas a decrease in the values was obtained from the heated and smoked samples. The increases observed in the heated samples were in agreement with the studies of Hamm (1966), who was of the opinion that changes in pH and free sulfhydryl groups were related to the unfolding of the peptide chains of myofibrillar proteins. The decreases observed in the heated and smoked samples were probably due to smoke constituent interactions with the various reactive groups within the proteins.

Table 2—A comparison of the pH, free sulfhydryl groups and the myofibrillar protein nitrogen content of untreated, heated and heated and smoked pork samples.[1]

State of muscle	pH	Variables: Sulfhydryl groups (μmoles/g protein)	Variables: Myofibrillar content (% of total nitrogen)
Heated	5.48	120.37	60.33
Untreated	5.31	91.87	47.57
Heated-smoked	4.95	69.81	35.79

[1]Smokehouse conditions: 60°C (140°F), 45% R.H., 2.25 hr.

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